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Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

Attachment

Claims 17, 19, 21-24, and 26-29 are rejected under 35 U.S.C. 103(a) as being unpatentable over Taira et al (US 2004/0002077) in view of Srivastava (USPN 5,948,646) for the reasons of record set forth in the Office actions mailed 12-13-07 and 8-1-08 and as set forth below.

Applicant's arguments filed 12-1-08 have been fully considered but they are not persuasive. Applicant argues that the instant claims are not obvious in view of Taira and Srivastava because the instant claims differ in that the prior art of record does not teach an intervening DNA sequence between the sense and antisense regions, which intervening sequence comprises an antibiotic resistance marker gene specifically positioned between two loxP sites. Instead, according to Applicant, the prior art only teaches an antibiotic resistance gene as a discrete transcription unit in an expression vector backbone. Applicant also argues that no rationale has been provided as to why a person of ordinary skill would be motivated to make the specific design choice of an intervening and removable antibiotic resistant marker gene from the huge spectrum of possible design choices available.

The claims are drawn to compositions and methods for transcribing RNAi in cells comprising providing eukaryotic cells at least one molecule of a nucleic acid comprising sense and antisense sequences of RNAi placed under control of a single transcription promoter, which sense and antisense sequence are separated by an intervening sequence flanked by lox sites, and which intervening sequence comprises a transcription stop site and a gene encoding an antibiotic resistance marker which is

optionally neomycin, wherein the intervening sequence is framed at each end by a pair of lox sites, and which method comprises providing Cre to cells and the expressed Cre produces site specific recombination and removal of the intervening sequence, thereby generating a construct encoding self complementary RNAi molecule under control of a single transcriptional promoter for subsequent gene silencing.

Taira et al teach methods and compositions comprising introducing a nucleic acid (DNA) molecule into a eukaryotic cell, which nucleic acid molecule comprises an antibiotic resistance marker, and further comprises a sense and antisense sequence optionally under control of a single transcriptional promoter, which sense and antisense sequences are separated by a stop codon, and which nucleic acid sequence is flanked by lox sites, and which method comprises providing Cre to cells and the Cre produces site specific recombination elimination of the intervening sequence, whereby upon recombination and in the presence of Cre, or upon Cre expression, RNAi is formed in the cell (see abstract, paragraphs 0023-0044, figures 1, 3-7, 10, paragraphs 0117-0119, 0126, claims 14, 17, 18, 22, 25-27).

Taira et al also teach compounds and methods of expressing RNAi in cells using an siRNA expression system, wherein said system comprises loxP sequences, wherein the two loxPs are located so as to interpose the antisense code DNA or sense code DNA; and Taira also teaches that the two loxPs are optionally arranged so as to interpose a linker comprising a stop sequence. Without CRE protein, transcription from the promoter is terminated at the stop sequence in the linker portion, leading to the suppression of siRNA production. CRE protein induces the recombination between

loxPs to displace the stop sequence, leading to transcription of antisense and sense code DNAs to produce a stem-loop siRNA (see paragraphs 0030-0035, 0081, 0098, and 0118).

Srivastava (USPN 5,948,646) teach the use of antibiotic resistant genes in expression plasmids, including hygromycin and neomycin resistant genes, to enhance selection of cells containing these antibiotic resistant selection markers (e.g. to enrich for transfected from untransfected cells) (see bridging paragraph, col. 16-17).

Contrary to Applicant's arguments, it would have been obvious to one of ordinary skill in the art to design and synthesize nucleic acid (DNA) molecules for methods of expressing RNAi in cells, relying on the prior teachings of Taira and Srivastava comprising introducing a nucleic acid molecule comprising a sense and antisense sequence under control of a single promoter, which sense and antisense sequences are separated by a stop codon, and which nucleic acid sequence is flanked by lox sites, whereby in the presence of, or upon Cre expression, RNAi is formed in the cell, because the method of producing RNAi molecules using a single transcription site, interposed with a stop codon between the sense and antisense sequences were taught previously using the nucleic acid constructs previously taught by Taira.

One would have been motivated to insert a neomycin resistant gene into this construct in order to determine the level of transfection of the nucleic acid construct in a target cell, and to select for cells comprising this construct. One of ordinary skill in the art would have expected that, prior to recombination, cells comprising this construct would be selected in the presence of neomycin, therefore enriching transfected cell

populations, and neomycin resistant markers were well known in the art at the time the instant invention was made. One of ordinary skill in the art would have been motivated to devise such a construct comprising a sense and antisense sequences flanked by Cre sites, so that, upon recombination, an RNAi construct would be formed for target gene inhibition from the two, self complementary sequences expressed as a single molecule from transcription of the transfected nucleic acid construct.

Contrary to applicant's assertions, one would have been motivated to come up with the instantly claimed design choice, containing a removable (antibiotic resistant) marker sequence, because the elimination of this intervening marker sequence allows for a prompt determination of successful recombination events in the cell, along with the ability to perform a comparison of the inhibition of target gene expression by properly formed siRNA molecules (lacking a long intervening sequence encoding a marker gene) in a host cell. The removable marker sequence therefore allows for a built-in assay to test recombination frequencies of previously transfected host cells without having to perform more laborious assays, such as sequencing.

One of ordinary skill in the art would have expected that, upon homologous recombination in the presence of Cre, or upon Cre expression, the resulting RNAi molecules would properly assemble, and target gene inhibition can then be readily tested along with an assessment of successful recombination. In this way, one can readily discern whether target gene inhibition has occurred or not, but following proper siRNA formation. The intervening marker is only present if no recombination occurs. And so if target gene inhibition does not occur, and antibiotic resistance has been lost in

the host cell, lack of target gene inhibition is likely due to poor siRNA activity, not due to lack of siRNA formation. The intervening marker sequence therefore allows a ready comparison between recombination abilities and gene silencing abilities.

For these reasons, the instant rejection is maintained.